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BIOINDICATORS IN AIR POLLUTION RESEARCH--APPLICATIONS AND CONSTRAINTS

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Physical and chemical measurements of air pollutants provide a precise measure of pollutant exposure which is frequently used to estimate probable biological impacts. In contrast, a biological response to the exposure indicates if the exposure had a biological effect. The biological response integrates the pollutant exposure and the modifying factors of genotype, climate and edaphic conditions. There is a growing interest in developing and using bioindicators to detect early changes in plant performance. Bioindicators may be classified as either accumulators of the pollutant or reactors to the pollutant. The ultimate selection of a bioindicator depends on the pollutant and the ultimate use of the data. A bioindicator should : 1) provide a readily detectable response to the pollutant; 2) be easy and efficient to use; 3) be reaily related to the resonse of interest; and 4) have a distinctive syndrome not readily confused with other causes. Bioindicators clearly have value in assessing environmental problems, but, at most, they are only indicators of a problem. Corrective or mitigative actions require additional data.

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Bioindicators in Air Pollution Research -- Applications and Constraints

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ABSTRACT

Physical and chemical measurements of air pollutants provide a precise measure of pollutant exposure which is frequently used to estimate probable biological impacts. In contrast, a biological response to the exposure indicates if the exposure had a biological effect. The biological response integrates the pollutant exposure and the modifying factors of genotype, climate and edaphic conditions. There is a growing interest in developing and using bioindicators to detect early changes in plant performance. Bioindicators may be classified as either accumulators of the pollutant or reactors to the pollutant. The ultimate selection of a bioindicator(s) depends on the pollutant and the ultimate use of the data. A bioindicator should: 1) provide a readily detectable response to the pollutant; 2) be easy and efficient to use; 3) be readily related to the response(s) of interest; and 4) have a distinctive syndrome not readily confused with other causes. Bioindicators clearly have value in assessing environmental problems, but, at most, they are only indicators of a problem. Corrective or mitigative actions require additional data (e.g., air monitoring).

INTRODUCTION

When attempting to assess the impact of an air pollutant(s) on individual organisms/species or whole ecosystems, the following questions are frequently posed:

"What is the current status of the species or ecosystem?"

"What is the current trend in the status of the species or ecosystem?"

These questions imply that there is a spatial or temporal change in the status of the system that has been or can be measured. The implied hope is that the data are currently available to conduct the assessement and that no additional studies are required to provide a timely and precise answer. However, if the data are not available, the issue becomes, "What physical or biological methods are readily available to conduct the assessment?

CLASSIFICATION OF ASSESSMENT METHODS

Physical/Chemical Methods: Physical/chemical methods (Guderian et al., 1985) can accurately describe, in "real time", the ambient

exposure at a given site or series of sites. These methods are sensitive, specific and reproducible but require electrical power making it difficult to situate monitors in remote locations. Also monitors are generally expensive to purchase and operate, requiring trained personnel. If resources and funds are available, monitors can be located throughout the study area permitting a more detailed characterization of exposure. Despite providing an accurate description of the exposure, these methods fail to consider the influence of other important factors that control biological responses. Although physical/chemical methods can establish the potential for biological impacts, they do not provide sufficient information to establish the risk of an exposure.

Biological Methods: A biological method is any response to a stress by an organism or community of organisms whose presence or other easily measured attribute can be assessed either qualitatively or quantitatively. Biological methods include bioindicators show the presence or absence of the stress and biomonitors which attempt to provide additional information about the intensity of the stress (Arndt et al., 1987; Guderian et al., 1985, Knabe, 1982). In contrast to physical/chemical methods, a biological response can not provide a precise measure of exposure. The biological response is influenced, not only by the concentration and duration of exposure to air pollutant(s) but also by the relative sensitivity of the organism. The relative sensitivity is a function of the stage of plant development, the previous and current edaphic and climatic conditions and the genetic composition of the plant. The unique advantage of biological response is that it integrates the influence of the various factors that control biological response; consequently they permit the direct assessment of risk from an exposure (i.e., they show if a given exposure(s) was harmful). Biological methods are frequently inexpensive and data can be obtained by periodic (e.g., monthly or yearly) visits to the sites.

The above comparisons of the various types of methods available for risk assessment lead to the obvious conclusion: there is no better indicator of the status of a species or a system than the species or system itself (i.e., biological methods). Biological methods integrate the effects of pollutant exposure and modifying factors. Consequently, biological methods should play a significant role in assessing the status of a species or ecosystem over temporal or spatial gradients, provided suitable measures of response can be found and properly applied (Guderian et al., 1985; Knabe, 1982).

CLASSIFICATION OF BIOLOGICAL METHODS

Biological methods may be classified (Arndt, et al., 1987; Guderian et al., 1985) as either passive or active (Figure 1). Passive methods use plants growing naturally in the study area, where they are ecologically adapted. However, with <u>in situ</u> vegetation it is difficult to partition the variation in response among individuals between that caused by different exposures and

that caused by genotypic variation. Also the specific responses of many species to specific pollutants have not been established, rendering a definitive diagnosis difficult. Despite these limitations, visible injury to vegetation has repeatedly provided the first indication of pollutant impacts. In contrast, active biological methods use standardized plants (known genotype and response) placed at specific locations to detect the presence of air pollutants. Bel W3 tobacco has been used extensively in North American and Europe as an active indicator to detect and confirm the widespread occurrence of photochemical oxidant air pollutants (Guderian et al., 1985).

An organism may be classified as either a reactor to or an accumulator of the pollutant (Figure 1). A reactor displays a typical symptom or a measurable response to the pollutant. Any measurable response of an organism can be used, however, foliar injury is probably used most frequently. To be a suitable bioindicator, the response must of an organism be specific for a particular pollutant and not readily confused with other similar symptoms with different causes. In contrast to a reactor, an accumulator will not necessarily display an overt symptom but will accumulate the pollutant causing in a significant tissue enrichment. Accumulators are only suitable for pollutants (e.g., fluorides or heavy metals) that have a long residence time in the tissue or for which the metabolic products are known.

A distinction is frequently made between using organisms as bioindicators or biomonitors (Figure 1). A bioindicator is an organism or biological response that reveals the presence or absence of an air pollutant by the occurrence of typical symptoms or measurable responses. A biomonitor provides information on the presence of the pollutant and attempts to provide additional information about the amount or intensity of the exposure. The bioaccumulation of pollutants has been proposed as a biomonitor. However, it is difficult to relate tissue concentrations of a pollutant to a specific concentration and/or duration of exposure because the tissue levels are influenced by the temporal nature of the pollutant occurrence as well as the edaphic, climatic and biological factors. In general, plants exposed to low concentrations absorb higher tissue levels than plants exposed to high concentrations that readily injure and kill the tissue. At this time, bioindicators are clearly important and have a history of use. However, biomonitors are not sufficiently defined and calibrated with respect to exposure to be reliable.

BIOINDICATORS

In various forms, bioindicators have been used almost as long as we have been aware of air pollution problems and there appears to be a renewed interest in using bioindicators to solve environmental problems. The primary goal of environmental protection is to protect humans, animals, plants and materials from adverse effects, not to reduce emissions or ambient concentrations. Bioindicators have a clear advantage over physical/chemical monitors because they indicate the status of

the organism.

A broad range of possible plant responses could potentially be used as bioindicators (Table 1). Because plants and ecosystems function across a wide range of scales of time and space, a diverse array of responses ranging from cellular processes (i.e., photosynthesis) to ecosystem responses (i.e., community changes) can be used to assess the state of the system.

The selection of a bioindicator(s) should consider several factors:

- o be easily measured and describe responses of concern within the ecosystem.
- o have a distinct response which is capable of predicting how the species/ecosystem will respond to the stress.
- o measure the response with acceptable accuracy and precision.
- o be based on a knowledge of the pollutant and its characteristics.
- o consider the final use of the data.

Although not a requirement, bioindicators are frequently selected to characterize responses of concern to humans. The bioindicator may be the response itself or related to the response of interest. For example, people are frequently interested in forest productivity but its measurement is a difficult process that is time consuming and can be expensive and uncertain. Consequently, a bioindicator or a surrogate for productivity, such as mid-summer leaf starch or leaf duration may be used to rapidly assess the status of the system. The hope is that these indicators will have sufficient accuracy and can rapidly and more easily be measured than productivity.

The selection of a bioindicator must consider the intended use of the data (Knabe, 1982). Will the data be used to determine the status of a species or the status of a community or ecosystem? A single bioindicator is not suitable for providing information at all levels of biological organization. The bioindicator must be appropriate for the different scales of time and space at which plants and ecosystems operate. Is the bioindicator being used to determine if there is an environmental problem in an area or to establish the bounds of a known problem? What level of accuracy and precision is needed? There is no reason to use a method yields greater precision and accuracy than needed for the intended use of the data. Will the bioindicator be used to assess the effects of air pollutants in the field or a laboratory study?

Knowledge of the particular air pollutant(s) is a principal consideration in the choice of a bioindicator(s). Not all bioindicators are appropriate for assessing the impacts of all air pollutants (Knabe, 1982).

EXAMPLES OF BIOINDICATORS AND THEIR LIMITATIONS:

Foliar symptoms are probably the most widely used bioindicator. There are numerous publications documenting the symptoms caused by specific air pollutants on a wide variety of plant species (e.g., Jacobson and Hill, 1970; van Haut and Strattman, 1970; Manning and Feder, 1980). The presence or absence of foliar injury has been used to establish zones of impact, while the type of foliar injury has been used to discriminate among various possible air pollutants. For example, foliar symptoms of ozone injury have been used extensively in field studies to establish the presence of ozone and the extent of its impact. These applications are based on the assumption that ozone produces a unique set of foliar symptoms which are not produced by other air pollutants or stressors. This assumption does not hold; mixtures of ozone and sulfur dioxide yield symptoms that are indistinquishable from those produced by ozone alone (Tingey et al., 1973). This problem is highlighted by the fact that publications describing symptoms of air pollutant injury also describe mimicking symptoms (i.e., symptoms induced by other stress factors which are readily confused with known air pollutant symptoms). Another limitation of foliar symptoms is that not all species have been exposed to all known air pollutants to establish their symptom expression. Consequently, it is still an art to diagnose the probable air pollutant on an untested species or genotype in the field. Additional data such as symptoms on adjacent (previously documented) species/genotypes or the availability of air monitoring data displaying elevated concentrations of the suspected air pollutant in the proper temporal sequence are required.

Elevated tissue concentrations of various elements has been extensively used to establish the presence of various air pollutants (Arndt et al., 1987). When chemical analysis is used, it is especially important to have some knowledge about the indentity of the suspected air pollutant and its possible source(s). For example if one has no knowledge about the probable air pollutant, it is very difficult to analyze plant tissue to determine if there are abnormal concentrations or accumulations of any inorganic element or organic compound in the tissue. These broad ranging analyses are very expensive and require large amounts of tissue and numerous extraction and analytical techniques in the hope of finding a elevated levels of an element or compound that can be judge to be a pollutant. Even if this broad spectrum analysis fails to identify a single compound, one can not dismiss the cumulative impact of several elements/compounds as a possible problem. However, when the suspected source of the pollutant or the likely chemical contaminant is known, it is much easier to pose focused questions, e.g., "Are fluoride levels elevated in the tissue?" "Is the tissue enriched in cadmium?" When the probable source is known, the optimum sampling design and analytical techniques can be chosen to determine the possible degree of contamination.

Tissue concentrations of various materials have been used as both passive and active bioindicators to map the concentrations of fluoride and various metals around pollution sources (e.g., Arndt, et al., 1987; Tingey et al., 1979). Once the concentration zones are mapped, then other biological responses, such as increased peroxidase activity, can be associated with various levels of the toxicant (Keller and Schwager, 1971). Tissue concentrations integrate the total exposure at the site, but it difficult to use this method as a biomonitor. Although tissue concentrations are dependent on the pollutant of exposure, the extent of tissue enrichment is influenced by several other factors, such as the location of the tissue on the plant, the plant developmental stage, environmental conditions and metabolic vigor of the organism. Plants absorb higher tissue levels when exposed to low concentrations than to high concentrations that injure the tissue. Enriched tissue concentrations alone is not a sufficient proof that a pollution problem exists. Additional analyses must establish that the tissue enrichment resulted from contamination by an anthropogenic source.

The identification of a proper control is critical when bioindicators are used to establish changes in plant performance over gradients in time or space in the field. This problem is highlighted by several examples. Air pollutants and other chemical stresses, as well as various abiotic and biotic stresses stimulate increased ethylene evolution by vegetation (Tingey 1980). This observation has been used to develop plant bioassays for a broad range of chemicals (Tingey 1980). However, it is doubtful if this approach can be used in the field because of the difficulty in establishing normal and abnormal ethylene emission rates. Also this elevated evolution of ethylene is a transient response. Similar difficulties exist when using plant pigment concentrations or a broad range of physiological or metabolic responses. Even the measure of plant growth suffers from the problem of establishing the appropriate control.

Care must be used in the interpretation bioindicator data in air pollution studies. In most cases cause and effect and specificity of the response have not been established. For example, ozone has been shown to induce the formation of hydroxyl radical adducts in chloroplast DNA (Floyd, personal communications). However, the stability of the adducts has not been determined and it is not known if the adducts are formed by other stresses. Consequently, the use of this response as a bioindicator is very limited. Similar types of problems limit the application of most bioindicators.

Bioindicators provide at most correlative responses that are not sufficient to instigate mitigative actions. A bioindicator response is analogous to having an elevated fever and going to the doctor. The fever is an indication of system dysfunction; it does not tell you what the disease is. Additional information and tests are required before the doctor can diagnose and propose a cure. So it is with bioindicator responses; a response is indicative of a potential problem but additional studies are

required to establish the problem and its magnitude. Only after the cause of the problem is established can specific mitigative or restorative actions be taken. Both physical/chemical and bioindicator methods have unique attributes and a combination of both approaches is necessary to conduct an adequate environmental assessement.

CONCLUSIONS:

Because bioindicators integrate the influence of environmental factors and provide direct measures of phytotoxicity of exposure(s) (i.e., risk), they clearly provide desirable information for environmental assessment. However, before they are used several factors must be established.

- o A bioindicator should provide readily detectable responses to the pollutant in the organism or the system of interest.
- o A bioindicator should be easy and efficient to use.
- o It is not necessary that there be a cause and effect association but the bioindicator must be a reliable predictor of the status of the organism or system.
- o A bioindicator should produce distinctive symptoms that are not readily confused with mimicking symptoms produced by other environmental stresses.
- o Both biological response and physical/chemical monitoring data are required for an environmental assessment program.

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Table 1. Examples of possible plant responses that could be used as bioindicators.

Plant Biomass
Growth/yield
Tree ring analysis
Morphological Responses
Foliar symptoms
Cellular/ultrastructural changes
Physiological Responses
Photosynthesis
Respiration
Stable isotope ratios
Plant Water Relations
Transpiration
Water potential and its components
Membrane permeability
Enzyme Activity
Tissue Analysis
Metabolite pools
Pollutant concentrations
Plant Pigments
Volatile Emissions
Bark Turbidity

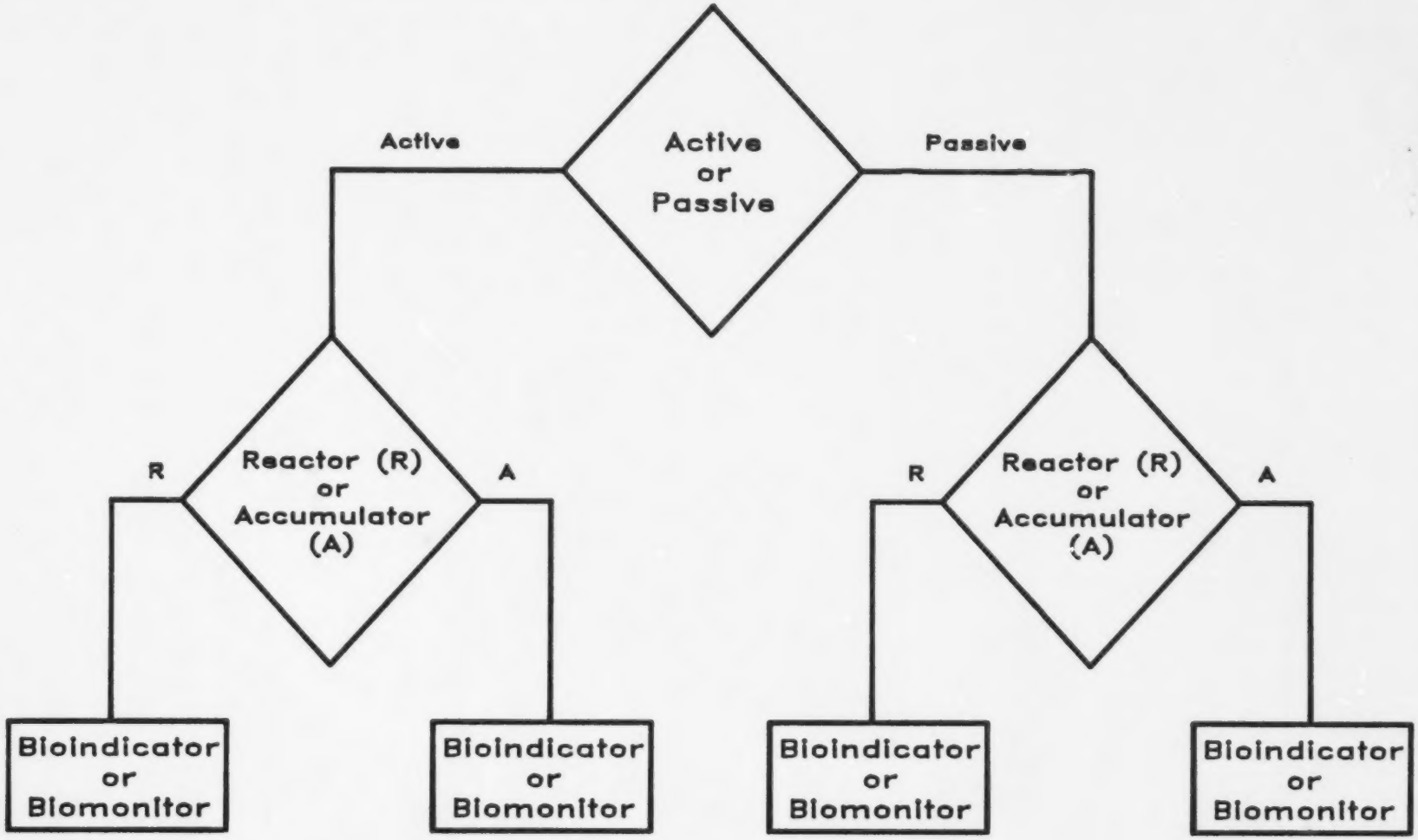

FIGURE LEGEND

Figure 1. A classification of biological methods used in air pollution studies.